percent potassium bromide disc prepared as described in paragraph (b)(1) of that section, except prepare a solution containing 3 milligrams of aztreonam per milliliter of methanol and use 0.5 milliliter of the solution as the sample.

[52 FR 4614, Feb. 13, 1987, as amended at 55 FR 11584, Mar. 29, 1990]

§455.10 Chloramphenicol.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity.* Chloramphenicol is a white to grayish-white or yellowish-white powder, occurring as needles or elongated plates. It is neutral, slightly soluble in water, but freely soluble in alcohol. It has the chemical formula D-(—)-*threo*—1-*p*—nitrophenyl-2 - dichloracetamido-1,3-propanediol. It is so purified and dried that:

(i) Its potency is not less than 900 micrograms per milligram.

(ii) [Reserved]

(iii) Its pH in a saturated aqueous solution is not less than 4.5 nor more than 7.5.

(iv) Its specific rotation in absolute ethyl alcohol at 20° C. is +20°±1.5°, and at 25° C. is +18.5°±1.5°.

(v) Its melting range is 151°±2° C.

(vi) Its absorptivity at 278 nanometers is 100 ±3 percent of that of the chloramphenicol working standard similarly treated.

(vii) It is crystalline.

(2) *Labeling.* It shall be labeled in accordance with the requirements of §432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of §431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for potency, pH, specific rotation, melting range, absorptivity, and crystallinity.

(ii) Samples required: 10 packages, each containing approximately 500 milligrams.

(b) *Tests and methods of assay*—(1) *Potency.* Use either of the following methods; however, the results obtained from the microbiological turbidimetric assay shall be conclusive.

(i) *Microbiological turbidimetric assay.* Proceed as directed in §436.106 of this chapter, preparing the sample for assay as follows: Dissolve an accurately weighed portion of the sample in sufficient 95 percent ethyl alcohol to obtain a solution containing 10,000 micrograms of chloramphenicol per milliliter (estimated). Add sufficient distilled water to obtain a concentration of 1,000 micrograms of chloramphenicol per milliliter (estimated). Further dilute an aliquot of the stock solution with distilled water to the reference concentration of 2.5 micrograms of chloramphenicol per milliliter (estimated).

(ii) *Spectrophotometric method.* Dissolve approximately 50 milligrams each of the sample and working standard in 100 milliliters of distilled water. Warm if necessary to hasten dissolution. Transfer 10 milliliters into a 250-milliliter volumetric flask and fill to volume with distilled water. Using a suitable spectrophotometer equipped with a 1-centimeter cell and distilled water as the blank, determine the absorbance of each solution at 278 nanometers. Calculate the potency of chloramphenicol as follows:

$$\text{Potency of sample in micrograms per milligram} = \frac{\text{Absorbance of sample} \times \text{weight of standard in milligrams} \times \text{potency of standard in micrograms per milligram}}{\text{Absorbance of standard} \times \text{weight of sample in milligrams}}$$

(2) [Reserved]

(3) *pH.* Proceed as directed in §436.202 of this chapter, using a saturated aqueous solution.

(4) *Specific rotation.* Accurately weigh approximately 1.25 grams of the sample into a 25-milliliter glass-stoppered volumetric flask and dissolve in about 15

milliliters of absolute alcohol, warming if necessary. Dilute the solution to 25 milliliters with absolute alcohol and mix thoroughly. Proceed as directed in § 436.210 of this chapter, using a 2.0-decimeter polarimeter tube.

(5) *Melting range.* Proceed as directed in § 436.209 of this chapter.

(6) *Absorptivity.* Proceed as directed in paragraph (b)(1)(ii) of this section, except calculate the percent relative absorptivity as follows:

$$\text{Percent relative absorptivity} = \frac{\text{Absorbance of sample} \times \text{weight of standard in milligrams} \times \text{potency of standard in micrograms per milligram}}{\text{Absorbance of standard} \times \text{weight of sample in milligrams} \times 10}$$

(7) *Crystallinity.* Proceed as directed in § 436.203(a) of this chapter.

[39 FR 19166, May 30, 1974, as amended at 45 FR 16476, Mar. 14, 1980; 48 FR 3960, Jan. 28, 1983; 50 FR 19921, May 13, 1985]

§ 455.10a Sterile chloramphenicol.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity.* Sterile chloramphenicol is a white to grayish-white or yellowish-white powder, occurring as needles or elongated plates. It is neutral, slightly soluble in water, but freely soluble in alcohol. It has the chemical formula D - (—) - *threo* - 1 - *p*—nitrophenyl - 2 - dichloracetamido - 1,3 - propanediol. It is so purified and dried that:

(i) Its potency is not less than 900 micrograms per milligram.

(ii) It is sterile.

(iii) It is nonpyrogenic.

(iv)—(v) [Reserved]

(vi) Its pH in a saturated aqueous solution is not less than 4.5 nor more than 7.5.

(vii) Its specific rotation in absolute ethyl alcohol at 20° C. is +20°±1.5°, and at 25° C. is +18.5°±1.5°.

(viii) Its melting range is 151°±2° C.

(ix) Its absorptivity at 278 nanometers is 100 ±3 percent of that of the chloramphenicol working standard similarly treated.

(x) It is crystalline.

(2) *Labeling.* It shall be labeled in accordance with the requirements of § 432.5(b) of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for potency, sterility, pyrogens, pH, specific rotation, melting range, absorptivity, and crystallinity.

(ii) Samples required:

(*a*) For all tests except sterility: 10 packages, each containing approximately 500 milligrams.

(*b*) For sterility testing: 20 packages, each containing approximately 50 milligrams.

(b) *Tests and methods of assay*—(1) *Potency.* Use either of the following methods; however, the results obtained from the microbiological turbidimetric assay shall be conclusive.

(i) *Microbiological turbidimetric assay.* Proceed as directed in § 436.106 of this chapter, preparing the sample for assay as follows: Dissolve an accurately weighed portion of the sample in sufficient 95 percent ethyl alcohol to obtain a solution containing 10,000 micrograms of chloramphenicol per milliliter (estimated). Add sufficient distilled water to obtain a concentration of 1,000 micrograms of chloramphenicol per milliliter (estimated). Further dilute an aliquot of the stock solution with distilled water to the reference concentration of 2.5 micrograms of chloramphenicol per milliliter (estimated).

(ii) *Spectrophotometric method.* Dissolve approximately 50 milligrams each of the sample and working standards in 100 milliliters of distilled water. Warm if necessary to hasten dissolution. Transfer 10 milliliters into a 250-milliliter volumetric flask and fill to volume with distilled water. Using a suitable spectrophotometer equipped with a 1-centimeter cell and distilled water